

Nutrient and fatty acid composition of the flesh of oil sardine (*Sardinella longiceps*) and Indian mackerel (*Rastrelliger kanagurta*) from Hadhramout coast of the Arabian Sea, Yemen

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Abstract

The current study was conducted to evaluate the nutritional characteristics (moisture, protein, lipids, ash and fatty acid composition) of the flesh of oil sardine (*Sardinella longiceps*) and Indian mackerel (*Rastrelliger kanagurta*) caught from Hadhramout coast of the Arabian Sea. The protein content was 21.6% and 18.1% (wet weight basis) for mackerel and sardine, respectively. The lipid content was much higher in sardine (10.1%) compared with mackerel (1.7%). The fatty acid composition showed that total saturated fatty acids had the highest relative value (37.5%) among other fatty acid groups in the flesh lipids of sardine, followed by polyunsaturated fatty acids (29.9%) and monounsaturated fatty acids (23.4%). In mackerel, polyunsaturated fatty acids was present at 37.4%, followed by saturated fatty acids (36.7%) and then monounsaturated fatty acids (14.3%). The majority of polyunsaturated fatty acids in both fish were deposited as omega-3 (89.8% in sardine and 87.9% in mackerel), of which docosahexaenoic acid and eicosapentaenoic acid were the most abundant. In conclusion, oil sardine and Indian mackerel which are locally available and affordable fish in Yemen can be considered valuable sources of nutrients particularly protein and health-beneficial omega-3 long chain polyunsaturated fatty acids.

Keywords

Nutrient composition
Arabian sea
Omega-3 fatty acids
Oil sardine
Indian mackerel

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Introduction

Fish have long been recognized as an important component of the diet of humans providing nutrients needed by the human body to function properly (Pigott and Tucker, 1990). The high nutritional benefits of fish consumption are mainly due to their proteins and lipids of high biological value, with long-chain polyunsaturated fatty acids (LC-PUFA), as well as certain minerals and vitamins (Sidhu, 2003). The nutrient content of fish varies greatly among species and from an individual fish to another depending on age, sex, environment, feed intake and season (Huss, 1995). Fish muscle usually contains 16 to 21% protein (Murray and Burt, 1969; Huss, 1995). Fish proteins are of high quality containing all of the essential amino acids in good quantity and in balanced amounts and are easily digested with digestibility values of greater than 90% (Pigott and Tucker, 1990). Fish lipids are characterized by its high content of omega-3 LC-PUFA such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). These fatty

acids are generally found in all fish but with higher concentrations in marine species with those from high latitudes having higher amounts than tropical low-latitude species (Cahu *et al.*, 2004; Garrido *et al.*, 2008; Huynh and Kitts 2009). The nutritional and health benefits of the consumption of fish and / or fish oil containing omega-3 polyunsaturated fatty acids (omega-3 PUFA) have been well documented. There is now a consensus that these omega-3 fatty acids particularly EPA and DHA have the ability to reduce the blood serum triglycerides, protect against cardiovascular diseases especially the acute complications of coronary heart disease such as sudden cardiac death and play a vital role in the development and functions of the nervous system (brain), photoreception (vision) and the reproductive systems (Leaf and Weber, 1988; Simopoulos, 1991; Gustafsson *et al.*, 1992; Kris-Etherton *et al.*, 2002; Sidhu, 2003; Cahu *et al.*, 2004). It has also been suggested that the intake of even relatively small amounts of fish is a favourable indicator of reducing

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the risk of several digestive tract cancers, notably colon and rectal cancer (Fernandez *et al.*, 1999).

Yemen has rich fisheries resources, with a total production of 228,655 metric tons in 2012 (Ministry of Fish Wealth, 2015). There are currently about 65 commercially important species including large pelagic fish (principally yellowfin and longtail tuna, kingfish and queenfish), small pelagic fish (particularly Indian oil sardine and Indian mackerel), demersal fish (groupers, emperors, jacks and bream) and invertebrates (shrimp, lobsters, cuttlefish and sea cucumbers) (Ministry of Fish Wealth, 2012).

Oil sardine (Indian oil sardine, *Sardinella longiceps*) is the most landed fish in Yemen, with annual catch of about 56000 metric tons in 2012 representing approximately 25% of the country's total fish production (Ministry of Fish Wealth, 2015). Indian mackerel (*Rastrelliger kanagurta*) is also landed in commercial quantities which were estimated to be about 15000 metric tons in 2012 (Ministry of Fish Wealth, 2015). These small pelagic fishes are generally available 'fresh' throughout the year with low prices and are commonly consumed in local communities. Both fish species are also utilized in some local industries such as fish canning and the fish meal and fish oil industry. Hence, primary information on the nutritional composition of these fishes is essential for food sources and industrial utilization. To the best of our knowledge such information is not available for oil sardine and Indian mackerel caught from the Yemeni coast of the Arabian Sea. The current study aimed to evaluate and compare the nutritional quality of oil sardine and Indian mackerel by providing data on the proximate and fatty acid compositions of these two fish species which constitute among the most abundant and low-cost fish in the Yemen region.

Materials and Methods

Collection and preparation of samples

The fish used in this study were obtained fresh from the central fish market in Mukalla, Hadhramout, Yemen. About 100 g flesh tissues were sampled from individual 10 fish of each species. The flesh samples from each species were divided into two sub-samples (five fish each). Flesh tissues for each sub-sample were then homogenized, wrapped in polyethylene film, sealed within plastic zipper bags and kept frozen (-18°C) until used for subsequent analyses.

Proximate analysis

Moisture, crude protein and ash content were analyzed according to Association of Official

Analytical Chemists (AOAC) standard methods (AOAC, 1997). Briefly, moisture was determined by drying the samples in an oven at 105°C until constant weight. Crude protein was determined (micro Kjeldahl method) by digesting the samples with concentrated H₂SO₄ followed by alkali (40% NaOH) distillation and acid (0.1 N HCl) titration. Ash content was determined by dry ashing in porcelain crucibles in a muffle furnace at 550°C for 5 hours. Total lipids (crude lipids) were extracted from samples based on the procedure of Bligh and Dyer (1959).

Fatty acid analyses

Crude lipids were esterified into methyl esters by saponification with 0.5 N methanolic sodium hydroxide and transesterified with methanolic boron trifluoride (AOAC, 1997). Fatty acid methyl esters (FAMES) were then resolved and analyzed using a Shimadzu gas-liquid chromatography (GC- A14). The esters were separated in an OmegawaxTM 320 fused silica capillary column (30 m × 0.32 mm, L × ID, 0.25 μm film thickness) from Supelco, Bellafonte Park, USA. A SPL-14 injector with a split ratio of 100:1 was used. Injector port and detector temperatures were set at 250°C and 260°C respectively. The temperature program was an initial temperature of 150°C for 2 min, with increase rate of 3°C/min to a final temperature of 220°C and held at this temperature for 10 min. Fatty acids were identified relative to retention time of known standards (Supelco 37 component FAME mix; Supelco, Bellafonte, PA) and areas beneath the identified chromatographic peaks were calculated by integration. Individual fatty acid content was shown as a percentage of the sum of total fatty acids detected.

Statistical analysis

All analyses were conducted in duplicates, and results were expressed as mean values ± standard deviation (SD). Independent t-test was used to test statistical differences between the two species using the SPSS program, version 17.0 for Windows (SPSS Inc. Chicago, IL, USA). Differences between means were considered to be significant at a P-value < 0.05.

Results and Discussion

Proximate composition

The proximate composition of the flesh of the two fish species is given in Table 1. All nutrient components measured (moisture, protein, lipid and ash) were significantly different (P < 0.05) between the two species. Values of crude lipid and ash were higher in sardine (10.1% and 1.7%, respectively) than

Table 1. Proximate composition (% wet weight) of fish flesh¹

Species	Composition			
	Moisture	Protein	Lipid	Ash
Oil sardine	68.9 ± 0.9	18.1 ± 0.1	10.1 ± 1.1	1.7 ± 0.0
Indian mackerel	75.0 ± 1.3	21.6 ± 0.0	1.7 ± 0.0	1.4 ± 0.0
Significance ²	*	*	*	*

¹ Values were reported as means ± S.D. of duplicate groups of 5 fish (n = 10).

² * significant (P < 0.05).

those of mackerel (1.7% and 1.4%, respectively). The difference in lipid content between the two pelagic fish species was especially significant. The values for flesh moisture and protein were higher in mackerel (75.0% and 21.6%, respectively) compared with sardine (68.9% and 18.1%, respectively).

Gokoglu and Yerlikaya (2015) and Huss (1995) reported that the moisture content of the flesh of fish ranges between 70-80%. Our results are within this range and comparable to values obtained by Ali *et al.* (2013) who reported moisture contents of 64.3-69.7% and 72.8-76.6% for oil sardine and Indian mackerel, respectively, in Oman. The lipid fraction of fish is the component that shows the greatest variation (Huss, 1995). Lipid content can be as low as 0.5% in lean starved fatty fish and can reach over 20% in well-fed fatty species (Johnston *et al.*, 1994). In addition to the inherent species differences other factors that may also contribute to such variations are season, geographical location as well as variations in age and maturity within the same species (Pigott and Tucker, 1990). Sardines and mackerels are commonly classified as fatty and oil-rich fish (Akman, 1988; Sen, 2005). Our result of high lipid content (10.1%) in oil sardine is typical for this fish species. The highest lipid content that we could find in literature for oil sardine was 11.9% for fish caught from the Indian coast (Gopakumar, 1997). The lipid content for oil sardine collected from various regions along the Arabian sea varied between 4.4% to 8.5% (Liyanage *et al.*, 1989; Ramesh, 2005 as cited by Ravichandran *et al.*, 2012; Ali *et al.*, 2013; Palani-kumar *et al.*, 2014). In contrast, the character of being a fatty fish was not obvious in Indian mackerel caught off the coast of Yemen. Available data showed fluctuating lipid contents from as low as 0.7% to as high as 12.0% for this species caught from the regional areas of the Arabian sea including Oman (Ali *et al.*, 2013), Pakistan (Nisa and Asadullah, 2011), India (Palani-kumar *et al.*, 2014) and Sri Lanka (Liyanage *et al.*, 1989). The lower lipid content found in Indian mackerel from the present study is within the range of 0.7-4.2% reported for fish samples caught from

Oman (Ali *et al.*, 2013). It would seem that oil sardine may provide a more reliable source of lipids as energy and essential fatty acids compared to Indian mackerel in Yemen and surrounding regions. Further studies should be conducted to elucidate the cause of the widely fluctuating lipid content observed in mackerels caught in the Arabian sea.

The protein content of most fishes has been suggested to be in the range of 16 to 21% (Murray and Burt, 1969; Huss, 1995). More specifically the values ranged between 15.9-20.1% for oil sardine (Ali *et al.*, 2013; Palani-kumar *et al.*, 2014) and 15.1-22.4% for Indian mackerel (Nisa and Asadullah, 2011; Ali *et al.*, 2013; Palani-kumar *et al.*, 2014) which are in accordance with the values determined in the present study. The average content of ash in fish muscle had been reported to be between 0.5% and 1.8% (Sidwell, 1981). Our results are within this range and comparable to those of 1.5-1.6% and 1.5% found in oil sardine and Indian mackerel, respectively (Ali *et al.*, 2013). A slightly lower ash content was reported for sardine and mackerel at 1.2% by Palani-kumar *et al.* (2014) and at 0.9-1.4% in mackerel by Nisa and Asadullah (2011).

Fatty acid composition

The fatty acid compositions of fish flesh are shown in Table 2. There were significant differences between the two fish species for most fatty acids. The percentage of total monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) was significantly higher (P < 0.05) in the flesh lipids of mackerel than that of sardine, while no significant differences (P > 0.05) were found in the total relative saturated fatty acids content between the two species. The fatty acid profile of sardine showed that saturated fatty acids (SFA) had the highest value (37.5%) among the fatty acid groups, followed by PUFA (29.9%) and MUFA (23.4%). While in mackerel, PUFA at 37.4% was the most abundant group of fatty acids, followed by SFA (36.7%) and MUFA (14.3%). The majority of PUFA (89.8% in sardine and 87.9% in mackerel) were deposited as omega-3 PUFA. These values

Table 2. Fatty acid composition (% total fatty acids) of fish flesh¹

Fatty acid	Species		Significance ²
	Oil sardine	Indian mackerel	
12:0	< 0.1	< 0.1	-
14:0	7.0 ± 0.08	3.3 ± 0.03	*
16:0	25.5 ± 0.34	23.1 ± 0.29	*
18:0	4.9 ± 0.04	10.6 ± 0.07	*
Total saturates	37.5 ± 0.46	36.7 ± 0.38	-
16:1n-7	6.7 ± 0.02	3.9 ± 0.01	*
18:1n-9	12.8 ± 0.22	7.2 ± 0.10	*
18:1n-7	ND ³	2.3 ± 0.06	*
20:1n-9	1.6 ± 0.02	0.8 ± 0.04	*
22:1n-11	2.4 ± 0.04	0.1 ± 0.07	*
Total monoenes	23.4 ± 0.13	14.3 ± 0.08	*
18:2n-6	1.4 ± 0.02	1.7 ± 0.02	*
18:3n-6	0.3 ± 0.00	0.3 ± 0.00	-
20:3n-6	0.2 ± 0.00	0.2 ± 0.04	-
20:4n-6	0.9 ± 0.00	2.0 ± 0.04	*
22:5n-6	0.3 ± 0.05	0.4 ± 0.02	-
Total omega-6 PUFA	3.0 ± 0.03	4.5 ± 0.13	*
18:3n-3	0.6 ± 0.01	1.1 ± 0.01	*
18:4n-3	2.0 ± 0.05	0.7 ± 0.00	*
20:3n-3	0.1 ± 0.00	0.2 ± 0.00	*
20:4n-3	0.5 ± 0.01	0.3 ± 0.00	*
20:5n-3	8.3 ± 0.14	4.8 ± 0.02	*
22:5n-3	1.1 ± 0.06	1.1 ± 0.02	-
22:6n-3	14.3 ± 0.53	24.7 ± 0.31	*
Total omega-3 PUFA	26.9 ± 0.41	32.8 ± 0.32	*
Total PUFA	29.9 ± 0.43	37.4 ± 0.20	*
omega-3: omega-6	8.8 ± 0.06	7.3 ± 0.27	

¹ Values were reported as means ± S.D. of duplicate groups of 5 fish (n = 10).

² * significant (P < 0.05); - not significant (P ≥ 0.05).

represented 26.9% and 32.8% of the total fatty acids in sardine and mackerel, respectively. As shown in Table 2 most of the omega-3 PUFA in the flesh lipids of both fish was contributed by DHA and EPA at 84% in sardine (53.1% from DHA and 31.0% from EPA) and 89.7% in mackerel (75.1% from DHA and 14.5% from EPA). On the other hand, the omega-6 PUFA content was much lower in both fishes. These fatty acids only accounted for 3.0% of total fatty acids (10.2% of total PUFA) in sardine and 4.5% of total fatty acids (12.1% of total PUFA) in mackerel.

As for individual fatty acids (Table 2) palmitic acid (16:0) was by far the highest single fatty acid detected in sardine at 25.5%. This was followed by DHA (22:6n-3), oleic acid (18:1n-9) and EPA (20:5n-3) at 14.3%, 12.8% and 8.3%, respectively. Other fatty acids detected in considerable quantities in the flesh lipids of sardine were 14:0, 16:1n-7 and 18:0 at 7.0%, 6.7% and 4.9%, respectively. Whereas in mackerel, DHA and palmitic acid were the first and second most abundant fatty acid with 24.7% and

22.8% of total fatty acids, respectively. These were followed by 18:0 (10.6%), 18:1n-9 (7.2%), 20:5n-3 (4.8%), 16:1n-7 (3.9%) and 14:0 (3.3%).

In general, results of fatty acid composition of both oil sardine and Indian mackerel observed in the present study are in accordance with the fatty acid profile of marine fishes. The trend of fatty acid profile showed for mackerel samples where PUFA was the predominant group, followed by SFA and then MUFA is typical for marine fish (Osman *et al.*, 2001; Sahena *et al.*, 2009). Similar trend has been previously reported for Indian mackerel (Liyanage *et al.*, 1989; Osman *et al.*, 2001; Marichamy *et al.*, 2009; Ganga *et al.*, 2010). The fatty acid profile was dominated by SFA, followed by PUFA and MUFA which was observed in sardine samples in this present study has also been reported for the same species caught from Sri Lanka (Liyanage *et al.*, 1989). The abundance of omega-3 PUFA particularly DHA and EPA is generally the major characteristic that has given marine fishes increasing global attention due

to the health-beneficial properties of these LC-PUFA (Sahena *et al.*, 2009). This feature was clearly shown in the fishes studied in the present work making the findings of this study significant from the nutritional and health point of view. With regards to omega-6 PUFAs content in marine fish, the amount of these fatty acids is much lower than omega-3 PUFAs (Sahena *et al.*, 2009). Our results are consistent with this trend and similar to earlier studies that reported low content of omega-6 PUFAs that is less than 5% of total fatty acids in marine fish (Ratnayake *et al.*, 1989; Njinkoue *et al.*, 2002). The fatty acid composition of fish is known to differ significantly among species and within species according to diet (Bahurmiz and Ng, 2007; Wijekoon *et al.*, 2014), season and environmental variables (Dutta *et al.*, 1985; Halilöglu *et al.*, 2004; Hong *et al.*, 2015) and whether fish are wild or farm-raised (Fuentes *et al.*, 2010; O'Neill *et al.*, 2015). One aspects of this variation is the inconsistency in fatty acids that dominate the fatty acid profile of fish which has been observed even within the same fish species from the same study area (Liyanage *et al.*, 1989; Görgün and Akpinar, 2012; Bouriga *et al.*, 2014; Khitouni *et al.*, 2014). The high relative content of palmitic acid in sardine and DHA in mackerel shown in the present study has been previously reported by Liyanage *et al.* (1989) for oil sardine and Liyanage *et al.* (1989), Osman *et al.* (2007) and Ganga *et al.* (2010) for Indian mackerel. However, other studies have reported EPA as the most abundant fatty acid in oil sardine (Som and Radhakrishnan, 2013) and palmitic acid as the most abundant in Indian mackerel (Marichamy *et al.*, 2009; Nisa and Asadullah, 2011). It is well documented that palmitic acid and DHA are two major fatty acid in marine fish (Osman *et al.*, 2007; Sahena *et al.*, 2009; Bouriga *et al.*, 2014). Palmitic acid is very important in the biosynthesis processes being a key metabolite in fish and that its level did not influenced by diet (Ackman and Eaton, 1966). Additionally, palmitic acid may serve a role as structural components of phospholipids in cell membranes (Perez *et al.*, 1999). DHA, along with EPA, are the principal omega-3 LC-PUFA which contained in high levels in fish oil. These fatty acids originate in unicellular phytoplanktons and seaweeds and accumulate in fish (Ackman 1980; Sahena *et al.*, 2009). Arachidonic acid (20:4n6, AA) is another essential fatty acid for humans that cannot be synthesized by the human body at a rate to meet the metabolic needs, hence must be provided via dietary ingredients (Linko and Hayakawa 1996; NAS/NRC, 2005). In the present study, AA was detected in fish lipids but in low amounts in sardine (0.90%) and higher in mackerel (2.0%). It has been

reported that this fatty acid usually occurs only at low or negligible levels in the lipids of marine fish (Saito *et al.*, 1999; Osman *et al.*, 2001; Njinkoue *et al.*, 2002; Zhao *et al.*, 2010) which is in good agreement with our results. The omega-3/omega-6 PUFA ratio has been suggested as a good indicator in comparing the relative nutritional values of fish oils of different species (Pigott and Tucker, 1990). The omega-3/omega-6 ratio of 1:1 is considered to be optimal for nutritional purposes (Simopoulos, 1989) and the higher ratios have been quoted as an index of higher nutritional value (Zhao *et al.*, 2010). In the present study the higher content of omega-3 PUFA and the lower content of omega-6 counterparts in both fish, led to high omega-3/omega-6 ratio at 8.8 for oil sardine and 7.3 for Indian mackerel. These values are identical with the range of 5 to 10 that has been suggested for the omega-3/omega-6 ratios of marine fish (Ahlgren *et al.*, 1994).

When evaluating fish as a 'functional' healthy food both the quantity of lipid and its content of omega-3 LC-PUFA are important factors to be taken into consideration (Rahman *et al.*, 1995). The high lipid content of oil sardine (10.1%) reported in this study along with its high omega-3 content make this fish a good source for these functional fatty acids. Although the lipid content in Indian mackerel is low, its high relative content of omega-3 PUFA especially DHA supports its value as a good source for healthy marine lipids.

In conclusion, results from the current study have shown the potential of oil sardine and Indian mackerel as good dietary sources for nutrients particularly protein and lipids with high omega-3 LC-PUFA. Further studies are required to provide more detailed data on the nutritive values of these fishes especially the amino acid and mineral compositions as well as the seasonal variation of these components.

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